Application No. 10/790,746 Amendment dated July 21, 2011 Reply to After Final Office Action of April 21, 2011

AMENDMENTS TO THE CLAIMS

Please amend the claims as follows:

1, - 15, (Canceled)

- 16. (Currently amended) A method of measuring the amount of la,25-dihydroxy vitamin D in human serum using a competitive protein binding assay, comprising:
 - i) separating 25-hydroxy vitamin D from $l\alpha_{\lambda}$ 25-dihydroxy vitamin D by binding $l\alpha_{\lambda}$ 25-dihydroxy vitamin D in a sample of the human serum to a material that specifically-binds $l\alpha_{\lambda}$ 25-dihydroxy vitamin D and eluting $l\alpha_{\lambda}$ 25-dihydroxy vitamin D from said material to provide a measurement sample,
 - ii) measuring the displacement of a vitamin D derivative of formula (I) from an antibody that specifically binds $l\alpha$, 25-dihydroxy vitamin D by adding an amount of the measurement sample to a sample-an amount of the antibody having the vitamin D derivative of formula (I) bound thereto,

wherein:

R represents a 25-hydroxylated side-group of vitamin D_2 or of vitamin D_3 , and Y represents hydroxy: and

iii) correlating the measurement of displacement of the vitamin D derivative of formula (I) from said antibody by $1\alpha,25$ -dihydroxy- $1\alpha,25$ dihydroxy-vitamin D present in the measurement sample to a measurement of displacement of the vitamin D derivative of formula (I) from the antibody by a known quantity of the $1\alpha,25$ -dihydroxy $1\alpha,25$ -dihydroxy vitamin D to determine the amount of $1\alpha,25$ -dihydroxy vitamin D in the sample.

17. - 18. (Canceled)

- 19. (Previously presented) The method of claim 16, wherein said competitive protein binding assay is selected from the group consisting of an enzyme immunoassay, an enzyme-linked immunosorbent assay, a radioimmunoassay, an immunoradiometric assay, a luminescence assay, a fluorescence immunoassay and an immunofluorometric assay.
- 20. (Previously presented) The method of claim 16, wherein the method is a sandwich immunoassay, selected from the group consisting of immunoradiometric assay, IEMA/EIA, immunoluminometric assay and immunofluorometric assay.
- 21. (Previously presented) A kit for determining the concentration of lo, 25-dihydroxy vitamin D in a sample of human scrum by an immune-based competitive protein binding assay, comprising

Application No. 10/790,746

Amendment dated July 21, 2011

Reply to After Final Office Action of April 21, 2011

a standardized quantity of a solid vitamin D derivative of formula (I) or a standardized solution of a vitamin D derivative of formula (I),

wherein R represents a 25-hydroxylated side-group of vitamin D_2 or of vitamin D_3 , and Y represents hydroxy;

a standardized quantity of an antibody that specifically binds $1\alpha,25$ -dihydroxy vitamin D; and a known quantity of $1\alpha.25$ -dihydroxy vitamin D.

so that the displacement of the vitamin D derivative of formula (I) from said antibody as effected by the $1\alpha_s 25$ -dihydroxy vitamin D present in the measurement sample can be correlated to the displacement of the vitamin D derivative of formula (I) from said antibody as effected by the addition of a known quantity of the $1\alpha_s 25$ -dihydroxy vitamin D to determine the amount of $1\alpha_s 25$ -dihydroxy vitamin D present in human serum.

- 22. (Previously presented) The kit of claim 21, further comprising a material that can bind 1α ,25-dihydroxy vitamin D for separation of 25-hydroxy vitamin D from 1α ,25-dihydroxy vitamin D.
- 23. (Previously presented) The kit of claim 21, wherein said competitive protein binding assay is selected from the group consisting of an enzyme immunoassay, an enzyme-linked immunosorbent assay, a radioimmunoassay, an immunoradiometric assay, a luminescence assay, a fluorescence immunoassay and an immunofluorometric assay.

Docket No. 0756-0124P

Application No. 10/790,746 Amendment dated July 21, 2011 Reply to After Final Office Action of April 21, 2011

- 24. (Previously presented) The kit of claim 21, wherein said competitive binding assay is a sandwich immunoassay, selected from the group consisting of immunoradiometric assay, IEMA/EIA. immunoluminometric assay and immunofluorometric assay.
- 25. (Previously presented) The kit of claim 21 comprising a solid phase selected from the group consisting of a microtitration plate, another solid earrier, a microparticle, a polymeric material, and a cellulose.
- 26. (Previously presented) The kit of claim 19, in which the solid phase is a microparticle comprising agarose.
- 27. (Previously presented) The kit of claim 19, in which the solid phase is a magnetic microparticle.
- 28. (Previously presented) The kit of claim 22, in which the material that can bind 1α,25-dihydroxy vitamin D for separation of 25-hydroxy vitamin D from 1α,25-dihydroxy vitamin D is one suitable for packing into a chromatographic column or one that is provided in a chromatographic column.